Available online at www.sciencedirect.com
SCIENCE DIRECT
Toxicology 186 (2003) 255–259
TOXICOLOGY
www.elsevier.com/locate/toxicol

Cadmium exposure and trace elements in human breast milk

Ryumon Honda [a,*], Kenji Tawara [b], Muneko Nishijo [b], Hideaki Nakagawa [b], Kyoko Tanebe [c], Shigeru Saito [c]

[a] *Department of Hygiene, Kanazawa Medical University, 1-1, Daigaku, Uchinada, Ishikawa 920-0293, Japan*
[b] *Department of Public Health, Kanazawa Medical University, 1-1, Daigaku, Uchinada, Ishikawa 920-0293, Japan*
[c] *Department of obstructive and Gynecology, Faculty of medicine, Toyama Medical Pharmaceutical University, Toyama, Japan*

Received 21 June 2002; accepted 6 December 2002

Abstract

The interrelations of the seven elements, calcium (Ca), magnesium (Mg), sodium (Na), potassium (K), phosphorus (P), copper (Cu), zinc (Zn), and cadmium (Cd) in human breast milk were examined in Japanese mothers to clarify the effects of Cd exposure on these important elements for infant growth. Breast milk and urine samples were obtained from 68 mothers, aged 19–38 years, at 5–8 days postpartum. The concentrations were determined by inductively-coupled plasma atomic emission spectrometry for Ca, Mg, Na, K, P, by flame atomic absorption spectrophotometry for Cu and Zn, and by flameless atomic absorption spectrophotometry for Cd. Geometrical mean Cd concentrations were 0.28 (geometrical standard deviation = 1.82) μg/l in breast milk and 1.00 (1.93) μg/g creatinine in urine. Among the above elements only Cd concentration in breast milk was significantly correlated with urinary Cd concentration ($r = 0.451$, $P < 0.001$). Significant positive correlations were found between Cu and Ca ($r = 0.500$, $P < 0.001$), Cu and Mg ($r = 0.378$, $P < 0.01$), and Zn and Mg ($r = 0.355$, $P < 0.01$) in breast milk. Cd concentration in breast milk showed an inverse relationship with Ca concentration in breast milk ($r = -0.248$, $P < 0.05$). These results indicate that the Cd concentration in breast milk closely reflects Cd body burden, with increased Cd in breast milk possibly affecting Ca secretion in breast milk.

Keywords: Cadmium; Minerals; Human breast milk

1. Introduction

The development of cadmium (Cd) toxicity is known to be related to the zinc (Zn) and copper (Cu) status in the liver and kidney in animals (Sato and Nagai, 1989; Liu et al., 1992, 1994) and humans (Nordberg, 1978; Honda and Nogawa, 1987). In animals, complex antagonistic interactions between Cd, Cu and Zn are well established, in which metallothionein is known to play an important role (Roberts et al., 1973; Magos and Webb, 1978). In a previous study of pregnant rats exposed to Cd, mobilization of Cd and metallothionein in the liver to kidney and placenta was found at the same time when Cu and Zn of the

* Corresponding author. Tel.: +81-76-286-2211; fax: +81-76-286-9723.
E-mail address: ryumon@kanazawa-med.ac.jp (R. Honda).

0300-483X/03/$ - see front matter
doi:10.1016/S0300-483X(03)00002-7

liver were transported to their fetuses (Chan and Cherian, 1993). In lactation, Cd is thought to be transported from maternal plasma to mammary gland and secreted into breast milk as well as Cu and Zn. However, the interaction between Cd and Cu or Zn has not been clarified in breast milk. Major nutritional elements like calcium (Ca), magnesium (Mg), phosphorus (P), sodium (Na) and potassium (K) in breast milk are required for infant development and growth, but the effects of Cd on their metabolism have not been sufficiently investigated either. Especially Ca in breast milk is reported to come from maternal bone, because of a decrease in maternal bone density noted in the lactation period (Sower et al., 1993; Kalkwarf et al., 1999). On the other hand, bone is a target organ of Cd toxicity, and decreased bone density is found in the general population exposed to even low doses of Cd (Alfven et al., 2000; Honda et al., 1997). However, the effect of Cd exposure on Ca or other elements related to Ca secretion in human breast milk is also unsettled. Thus in this study we tried to clarify the interrelations between the major nutritional elements Ca, Mg, P, Na, K and the essential trace elements Cu and Zn, and Cd in human breast milk.

2. Materials and methods

2.1. Subjects

The subjects were 68 women aged 19–38 years, comprising 69.4% of the women who delivered their infants at Toyama Medical University Hospital and who had sufficient breast milk to provide samples after feeding their babies. The subjects were all Japanese and lived in non-industrial area close to the hospital, and more than 70% of them were housewives whose most common job before marriage was office work. There were no significant differences in nutritional status or health management practices between the subjects who participated in the present study and those who did not. Informed consent for this study was obtained from all subjects in the appropriate manner. Information about occupational exposure to Cd, residence in a Cd polluted area and smoking habit was obtained via a questionnaire filled out by the subjects themselves, and confirmed by maternity nurses during the hospital admission after delivery.

2.2. Analysis of urine and breast milk

Urine and transitory milk samples were collected into polypropylene tubes on the 5th postpartum day or 8th day in the cases with delayed milk excretion because of cesarean section. The samples were kept frozen at −20 °C until analyses. Cd concentration in urine was analyzed by flameless atomic absorption spectrophotometry after wet ashing in $HNO_3/H_2SO_4/HClO_4$ and extraction with APDC (ammonium pyrrolidine dithiocarbamate)-MIBK (methyl isobutyl ketone), and corrected for the urinary creatinine concentration. The concentrations were determined by inductively-coupled plasma atomic emission spectrometry for Ca, Mg, Na, K, P and by flame atomic absorption spectrophotometry for Cu and Zn, and by flameless atomic adsorption spectrophotometry for Cd, after digestion by $HNO_3/HClO_4$ and dilution with HNO_3.

2.3. Statistical analysis

Student's t-test or a Welch's test was used in the analysis of data to determine significant bivariate differences between groups with and without characteristics of mothers and delivery. Simple correlations between urinary Cd and seven other elements in breast milk, and between Cd and Mg, Na, K, Ca, P in breast milk were tested by Spearman's rho to determine the relationship between them.

3. Results

Cadmium concentration in breast milk of the subjects was 0.07–1.23, geometrical mean 0.28 (geometrical standard deviation = 1.82) μg/l. The levels of Cd in breast milk were not affected by maternal age, parity, or cesarian operation status (Table 1). The observed eight elements concentrations in breast milk of these subjects were shown in

Table 1
Comparisons of elements concentrations in breast milk between two groups according to maternal age and history of delivery

	No.	Elements in breast milk															
		Cd (µg/l)		Cu (µg/l)		Zn (mg/l)		Na (mg/l)		K (mg/l)		Mg (mg/l)		Ca (mg/l)		P (mg/l)	
		Mean# (min–max)	(S.D.)#	Mean# (min–max)	(S.D.)#	Mean (min–max)	(S.D.)	Mean# (min–max)	(S.D.)#	Mean (min–max)	(S.D.)	Mean (min–max)	(S.D.)	Mean (min–max)	(S.D.)	Mean (min–max)	(S.D.)
Maternal age																	
≥ 35	10	0.379 (0.17–1.22)	1.78	263.0 (119–516)	1.47	5.41 (3.39–7.99)	1.44	371.5 (183–750)	1.75	678.3 (480–762)	85.0	32.2 (19.4–41.2)	7.1	344.4 (256–451)	65.2	191.6 (136–280)	38.7
< 35	58	0.263 (0.07–1.06)	1.81	312.6 (123–705)	1.44	5.90 (2.73–11.6)	1.83	345.9 (189–1491)	1.47	727.8 (444–918)	75.5	34.7 (23.5–61.7)	6.9	326.4 (166–640)	79.3	188.6 (81.6–309)	45.7
Parity																	
Nullipara	35	0.277 (0.11–1.06)	1.94	309.0 (155–692)	1.45	6.27 (2.73–11.6)	1.92*	373.3 (224–674)	1.37	738.3 (582–918)	69.4	35.1 (24.0–61.7)	7.8	327.8 (185–640)	85.8	183.9 (102–309)	51.1
Multipara	33	0.278 (0.07–1.22)	1.72	295.1 (119–705)	1.46	5.35 (2.83–7.99)	1.50	327.3 (183–1491)	1.63	701.6 (444–822)	83.7	33.6 (19.4–43.8)	5.8	330.4 (166–521)	67.7	194.5 (81.6–280)	36.1
Caesarean operation																	
With	15	0.264 (0.15–1.22)	1.78	292.4 (119–692)	1.58	5.8 (3.72–8.62)	1.58	373.3 (183–1491)	1.84	701.9 (444–918)	116.1	35.5 (19.4–46.2)	6.6	327.7 (166–462)	90.5	183.6 (81.6–262)	53.3
Without	53	0.329 (0.07–1.06)	1.93	308.3 (128–705)	1.41	5.83 (2.73–11.6)	1.84	364.7 (193–750)	1.41	725.8 (582–855)	64.4	34.1 (23.5–61.7)	7.1	329.5 (220–640)	90.5	190.6 (102–309)	42.1
Total	68	0.277 (0.07–1.22)	1.82	304.8 (119–705)	1.45	5.32 (2.73–11.6)	1.78	340.0 (183–1491)	1.51	711.5 (444–918)	111.3	34.6 (19.4–61.7)	6.9	329.1 (166–640)	77.0	189 (81.6–309)	44.5

No.: number of subjects.
* $P < 0.05$ Significant difference as compared with the counterpart.
\# Geometrical mean and S.D.

Table 1. The level of Zn in breast milk of nulliparas was significantly higher than that of multiparas (Table 1). There was no difference in the concentrations of other elements in breast milk between the two groups according to these factors related to delivery.

To clarify the effect of Cd exposure on the major nutritional elements and trace elements in breast milk, the correlations between urinary Cd and Na, K, Ca, Mg, P, Cu, Zn and Cd in breast milk were computed. Among the eight elements in breast milk, only Cd concentration in breast milk was significantly correlated with urinary Cd concentration which is known to be an index of Cd body burden ($r = 0.451$, $P < 0.001$) (Table 2). Urinary Cd concentrations of these subjects were 0.28–5.13, (geometrical mean 1.00, geometrical standard deviation = 1.93) µg/g creatinine. Although the results were not shown in the tables, no significant relationship between Cd in breast milk and maternal age was found.

The interaction of metals and nutritional elements in breast milk is another focus of this study. Significant positive correlations were found between Cu and Ca ($r = 0.500$, $P < 0.001$), and Cu and Mg ($r = 0.378$, $P < 0.001$), and Zn and Mg ($r = 0.355$, $P < 0.01$) concentrations in breast milk. However, Ca concentration in breast milk showed a significant inverse relationship with Cd concentration in breast milk ($r = -0.248$, $P \leq 0.05$) (Table 3).

4. Discussion

Breast milk is not only an important source of various nutrients, but also a source of environmental pollutants, heavy metals and dioxins and so on. Although Cd in breast milk is one of the major sources of Cd exposure for infants, the effects of Cd in breast milk on infant health have not been well clarified, because the levels of Cd in breast milk are low, and no relationship between maternal Cd exposure and Cd in breast milk was found in previous European studies (Radish et al., 1987; Oskarsson et al., 1998). We previously reported that the Cd concentration in breast milk of mothers with high urinary concentrations (≥ 2

μg/g Cr) was higher than that of mothers with less urinary Cd (Nishijo et al., 2002). The level of Cd in breast milk in this study was higher than that reported in European countries, because Cd body burden is higher in Japan (WHO, 1992). In this study based on a greater number of subjects drawn from non-industrial area with a wide range of urinary Cd concentrations, it was confirmed that Cd in breast milk increased in parallel with increases in urinary Cd.

The levels of major nutritonal and trace elements except Cd in breast milk of these subjects were similar to those reported in previous reports in European or Asian countries (Arnaud and Favier, 1995; Lin et al., 1998; Wasowicz et al., 2001). However, the influences of maternal age and parity on these elements in breast milk were controversial in these studies. Lin et al. (1998) reported that Zn concentration in breast milk of mothers aged ≥ 30 years old was higher than that of younger mothers aged 20–29 years old, despite which they were unable to detect a significant relationship between Zn in breast milk and parity. In contrast, we could not find any differences in the Zn concentration between different aged groups in this study, and found that Zn levels in breast milk of nulliparas, who are younger than multiparas, was higher than that of multiparas. Similarly, Arnaud et al. (Arnaud and Favier, 1995) reported the effect of parity on Cu concentrations in breast milk, but no such effect was noted in the present subjects. These inconsistent results may be attributable to differences in lifestyle, especially dietary habits.

Table 2
Correlation coefficients between urinary Cd and eight elements in breast milk

Milk elements	Correlation coefficients
Cd	0.451***
Cu	−0.197
Zn	−0.066
Na	−0.117
K	−0.056
Ca	−0.073
Mg	−0.064
P	0.097

*** $P < 0.001$.

Table 3
Correlations between trace elements and major nutritional elements in breast milk

	Trace elements		
	Cd	Cu	Zn
Na	−0.052	0.066	0.066
K	0.093	0.146	0.224
Ca	−0.248*	0.500***	−0.013
Mg	0.027	0.378**	0.355**
P	0.049	0.049	−0.078

* $P < 0.05$,
** $P < 0.01$,
*** $P < 0.001$.

The most interesting issue in the present study is the interaction between Cd and major nutritional and trace elements in breast milk, because the nutritional function of milk is important for infant health. In the liver or kidney, the interactions of Zn and Cd or Cu and Cd are well known from the earlier literature (Nordberg, 1978; Honda and Nogawa, 1987), but we could not detect their relations in breast milk of these subjects. The differences in the secretion mechanisms, in which metallothionein is not involved, between Cd and Cu or Zn in breast milk need to be investigated in future.

In this study, Cd was negatively related to Ca in breast milk, whereas Zn and Cu were positively correlated with Ca or Mg. Early breast milk Ca was reported to be provided by maternal renal conservation of Ca and by loss of spinal tubular bone. These sites, renal tubules and bone, are well known target organs of Cd toxicity, even at low exposure levels (Alfven et al., 2000; Honda et al., 1997). Ohta et al. (1998) reported the effect of Cd on bone density and renal function in pregnant and lactating rats. These results suggest that Ca metabolism in bone and kidney are affected by Cd in humans.

Phosophorus also influences Ca uptake, and Ca/P ratio is important in bone mineralization in bone and Ca excretion in kidney, but no relationship was found between P, Ca/P and Cd in breast milk. There is another possibility of inhibition of Ca

transport by Cd in the mammary grand, because urinary Cd did not show a significant relation to Ca in breast milk.

More studies on the effects of Cd on bone mass and on intestinal Ca absorption of mothers are necessary to clarify the mechanism of decreased Ca by Cd exposure in breast milk. However, this relationship between Ca and Cd suggests that a high level of Cd in breast milk is a matter of not only additional body burden but also decreased Ca intake in the infants.

5. Conclusion

The present results indicate that Cd concentration in breast milk is closely associated with environmental Cd exposure, and that there may be an inverse relationship between Cd and Ca concentrations in breast milk.

Acknowledgements

We thank the nursing and medical staff at Toyama Medical Pharmaceutical University Hospital; Makiko Ebie, Mutsumi Katsuo, Aki Ota, and Dr Masatoshi Sakai. This work was supported by grant for project research from High Technology Center of Kanazawa Medical University (H2000-4).

References

Alfven, T., Elinder, C.G., Carlesson, M.D., Grubb, A., Hellström, L., Persson, B., Petersson, C., Spång, G., Schütz, A., Järup, L., 2000. Low-level cadmium exposure and osteoporosis. J. Bone Miner. Res. 15, 1579–1586.

Arnaud, J., Favier, A., 1995. Copper, iron, manganese and zinc contents in human colostrums and transitory milk of French women. Sci. Total Environ. 159, 9–15.

Chan, H.M., Cherian, M.G., 1993. Mobilization of hepatic cadmium in pregnant rats. Toxicol. Appl. Pharmacol. 120, 308–314.

WHO. Environmental Health Criteria 134, (1992) Cadmium. Geniva: WHO.

Honda, R., Nogawa, K., 1987. Cadmium, zinc and copper relationships in kidney and liver of humans exposed to environmental cadmium. Arch. Toxicol. 59, 437–442.

Honda, R., Tsuritani, I., Ishida, M., Noborisaka, Y., Yamada, Y., 1997. Relationship between ultrasonic calcaneal bone measurements and urinary cadmium excretion in women living in a non-Cd-polluted area in Japan. J. Bone Miner. Res. 12 (Suppl. 1), S1–S576.

Kalkwarf, H.J., Specker, B.L., Mono, H.O., 1999. Effects of calcium supplementation on calcium homeostasis and bone turnover in lactating women. J. Clin. Endocrin. Metab. 84, 464–470.

Lin, H.E., Jong, Y.J., Chiang, C.H., Yang, M.H., 1998. Longitudinal changes in Ca, Mg, Fe, Cu, and Zn in breast milk of women in Taiwan over a lactation period of one year. Biol. Trace Element Res. 62, 31–41.

Liu, X., Jin, Y., Nordberg, G., Rannar, S., Sjostrom, M., Zhou, Y., 1992. A multivariate study of protective effects of Zn and Cu against nephrotoxicity induced by cadmium metallothionein in rats. Toxicol. Appl. Pharmacol. 114, 239–245.

Liu, X., Jin, Y., Nordberg, G., Sjostrom, M., Zhou, Y., 1994. Influence of Zinc and copper administration on metal disposition in rats with cadmium-metallothionein-induced nephrotoxicity. Toxicol. Appl. Pharmacol. 126, 84–90.

Magos, L., Webb, M., 1978. Theorectical and practical considerations on the problem of metal–metal interaction. Environ. Health Perspect. 25, 151–154.

Nishijo, M., Nakagawa, H., Honda, R., Tanebe, K., Saito, S., Teranishi, H., and Tawara, K. (2002). Effects of maternal exposure to cadmium on pregnancy outcome and breast milk. J. Occup. Environ. Med. 59, 394–397.

Nordberg, G., 1978. Factors influencing metabolism and toxicity of metals: A consensus report by the task group on metal interaction. Environ. Health Perspect. 25, 3–41.

Ohta, H., Asami, S., Seki, Y., Yoshikawa, H., 1998. Effect of oral cadmium administration on the bone mineral density and renal function in female rats. Metal Ions Biol. Med. 5, 655–660.

Oskarsson, A., Palminhger, H., Sundbeg, J., Petersson, K., 1998. Risk assessment in relation to neonatal metal exposure. Analyst 123, 19–23.

Radish, B., Werner, L., Nau, H., 1987. Cadmium concentration in milk and blood of smoking mothers. Toxicol. Lett. 36, 147–152.

Roberts, K.R., Miller, W.J., Stake, P.E., Gentry, R.P., Neathery, M.W., 1973. High dietary cadmium on zinc absoption and metabolism in calves fed for comparable nitrogen balances. Proc. Soc. Exp. Biol. Med. 144, 906–908.

Sato, M., Nagai, Y., 1989. Effect of zinc deficiency on the accumulation of metallothionein and cadmium in the rat liver and kidney. Arch. Environ. Contam. Toxicol. 18, 587–593.

Sower, M., Corton, G., Shapiro, B., Jannaussh, M.L., Crutchfield, M.C., Smith, M.L., Randolph, J.F., Hollis, B., 1993. Changes in bone density with lactaion. J. Am. Med. Assoc., 269, 3130–3135.

Wasowicz, W., Gromadzinska, J., Szram, K., Rydzynski, K., Cieslak, J., Pietrzak, Z., 2001. Selenium, zinc, and copper concentrations in the blood and milk of lactating women. Biol. Trace Element Res. 79, 221–233.